STIC-Biotech/ChemLib 92825

From: Chan, Christina
Sent: Wednesday, April 30, 2003 11:07 AM
To: Davis, Minh-Tam; STIC-Biotech/ChemLib
Subject: RE: Rush search request for 09/030606

Please rush. Thanks Chris

Chris Chan
TC 1600 New Hire Training Coordinator and SPE 1644
308-3973
CM-1, 9B19

-----Original Message-----
From: **Davis, Minh-Tam**
Sent: Wednesday, April 30, 2003 9:49 AM
To: Chan, Christina
Subject: Rush search request for 09/030606

Please search:
1) SEQ ID NOs:110, 173,174,175,177, and 223 against sequences in US 2002/0086301 A1, US 6,252,027B1, 08/850713 and US 6,130043
2) SEQ ID NOs:110, 173,174,175,177, and 223 against sequences in the parent cases to determine priority: 09/020747, 08/904809, 08/806596.
Thank you.
MINH TAM DAVIS
ART UNIT 1642, ROOM 8A01, MB 8E12
305-2008

Point of Contact:
Toby Port
Technical Info. Specialist
CM1 6A04
703-308-3534

Searcher:_____
Phone:_____
Location:_____
Date Picked Up: 4/30
Date Completed: 5/1
Searcher Prep/Review:_____
Clerical:_____
Online time:_____

TYPE OF SEARCH:
NA Sequences:_____
AA Sequences:_____
Structures:_____
Bibliographic:_____
Litigation:_____
Full text:_____
Patent Family:_____
Other:_____

VENDOR/COST (where applic.)
STN:_____
DIALOG:_____
Questel/Orbit:_____
DRLink:_____
Lexis/Nexis:_____
Sequence Sys.:_____
WWW/Internet:_____
Other (specify):_____

0095719 99095361 PMID: 9879288

Expression of alpha-fetoprotein and prostate-specific antigen genes in several tissues and detection of mRNAs in **normal** circulating **blood** by reverse transcriptase-polymerase chain reaction.

Ishikawa T; Kashiwagi H; Iwakami Y; Hirai M; Kawamura T; Aiyoshi Y; Yashiro T; Ami Y; Uchida K; Miwa M

Department of Biochemistry and Molecular Oncology, Institute of Basic Medical Sciences and Center for Tsukuba Advanced Research Alliance, Ibaraki, Japan.

Japanese journal of clinical oncology (JAPAN) Dec 1998, 28 (12) p723-8, ISSN 0368-2811 Journal Code: 0313225

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

BACKGROUND: alpha-Fetoprotein (AFP) and prostate-specific antigen (PSA) in serum are widely used as tumor markers in the evaluation of prognosis and management of patients with hepatocellular carcinoma and prostate cancer, respectively. To establish the molecular diagnosis of cancer, reverse transcriptase polymerase chain reaction (RT-PCR) for AFP and PSA was used to identify circulating cancer cells in the blood of cancer patients. Here, we examined the tissue-specificity of AFP and PSA and tested whether AFP and PSA are suitable targets in the detection of certain cancer cells by RT-PCR using peripheral blood samples. METHODS: Tissue specificity of AFP and PSA was analyzed by Northern blotting and RT-PCR. Probes for AFP and PSA were hybridized with poly A+ RNAs from 50 human tissues. RT-PCR for AFP and **PSA mRNA** was performed using several cancerous tissues and **normal** tissues and peripheral **blood** cells from seven healthy volunteers. RESULTS: Broad expression of AFP was observed in several tissues and a large amount of AFP **mRNA** was found in fetal liver. **PSA** was expressed in prostate, salivary gland, pancreas and uterus. By RT-PCR, AFP and **PSA mRNA** were detected in several tumors, including salivary pleomorphic adenoma, hilar bile duct carcinoma, pancreatic carcinoma, transitional cell carcinoma of urinary bladder and thyroid papillary carcinoma. Furthermore, AFP and **PSA mRNAs** were frequently detected by RT-PCR, even in peripheral blood cells from healthy volunteers. CONCLUSIONS: Neither AFP nor **PSA** showed tissue-specific expression. AFP and **PSA mRNA** were detected in several diseased and non-diseased tissues and **normal** circulating **blood** by RT-PCR.

... alpha-fetoprotein and prostate-specific antigen genes in several tissues and detection of mRNAs in **normal** circulating **blood** by reverse transcriptase-polymerase chain reaction.

... were hybridized with poly A+ RNAs from 50 human tissues. RT-PCR for AFP and **PSA mRNA** was performed using several cancerous tissues and **normal** tissues and peripheral **blood** cells from seven healthy volunteers. RESULTS: Broad expression of AFP was observed in several tissues and a large amount of AFP **mRNA** was found in fetal liver. **PSA** was expressed in prostate, salivary gland, pancreas and uterus. By RT-PCR, AFP and **PSA mRNA** were detected in several tumors, including salivary pleomorphic adenoma, hilar bile duct carcinoma, pancreatic carcinoma, transitional cell carcinoma of urinary bladder and thyroid papillary carcinoma. Furthermore, AFP and **PSA mRNAs** were frequently detected by RT-PCR, even in peripheral blood cells from healthy volunteers. CONCLUSIONS: Neither AFP nor **PSA** showed tissue-specific expression. AFP and **PSA mRNA** were detected in several diseased and non-diseased tissues and **normal** circulating **blood** by RT-PCR.

4/3,K,AB/2 (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09740518 98171839 PMID: 9510850

Expression of prostate-specific antigen and prostate-specific membrane antigen transcripts in blood cells: implications for the detection of hematogenous prostate cells and standardization.

Gala J L; Heusterspreute M; Loric S; Hanon F; Tombal B; Van Cangh P; De Nayer P; Philippe M

Department of Biochemistry, Cliniques Universitaires Saint-Luc,

09740518 98171839 PMID: 9510850

Expression of prostate-specific antigen and prostate-specific membrane antigen transcripts in blood cells: implications for the detection of hematogenous prostate cells and standardization.

Gala J L; Heusterspreute M; Loric S; Hanon F; Tombal B; Van Cangh P; De Nayer P; Philippe M

Department of Biochemistry, Cliniques Universitaires Saint-Luc, Universite Catholique de Louvain, Bruxelles, Belgique. gala@sang.ucl.ac.be

Clinical chemistry (UNITED STATES) Mar 1998, 44 (3) p472-81, ISSN 0009-9147 Journal Code: 9421549

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Circulating prostate cells can be detected in cancer patients by using reverse transcriptase-PCR (RT-PCR) assay for prostate-specific antigen (**PSA**) and prostate-specific membrane antigen (PSM) **mRNA**. A quality-control study involving a conventional RT-PCR assay was performed and, surprisingly, detected both transcripts in many negative control cell lines and in **normal blood** samples. The existence of an illegitimate transcription of the PSA and PSM genes was evidenced by sequence analysis of several PSM and PSA-PCR products. Sequencing indeed demonstrated the presence of a PSA or PSM polymorphism in some but not all the cell lines and patient samples, as well as a heterozygous mutation (G to A; Asp to Asn) in the Jurkat cell line. Moreover, the amount of PSA transcript in MCF-7, a PSA-negative breast line, increased after incubation with cycloheximide. Interestingly, the frequency of positivity was as high as 12% in male samples if only tested once, but dropped to 3% upon multiple testing of the same cDNA. This highlights the stochastic effects in RT-PCR results at high sensitivity, hence the importance of repetitive testing in clinical samples. Decreasing the number of cycles avoided the amplification of illegitimate transcripts but also affected the limit of detection, as evidenced with PSA and PSM cDNA containing plasmids, mixing of LNCap with **normal blood** samples, and the PSA-PSM-negative K562 cell line.
The current data raise the need for a multicentric standardization of the RT-PCR methodology used to amplify PSA and PSM transcripts.

```
      ---  -----  -----------
? s psa (10n)mRNa??
          23843  PSA
         459790  MRNA??
      S1     460  PSA (10N)MRNA??
? s normal (5n) (blood or circulation)
        1796377  NORMAL
        3385902  BLOOD
        1058705  CIRCULATION
      S2   51673  NORMAL (5N) (BLOOD OR CIRCULATION)
? s s1 and s2
            460  S1
          51673  S2
      S3      12  S1 AND S2
? rd
>>>Duplicate detection is not supported for File 340.

>>>Records from unsupported files will be retained in the RD set.
...completed examining records
      S4       4  RD (unique items)
? t s4/3,k,ab/1-4

 4/3,K,AB/1     (Item 1 from file: 155)
DIALOG(R)File 155:MEDLINE(R)
```

```
Set     Items   Description
S1      18595   PROSTATE(2N)CELL??
S2    4386413   SERUM OR BLOOD OR CIRCULATION
S3       3236   S1 AND S2
S4    2184502   NORMAL OR HEALTHY
S5        803   S3 AND S4
S6    2314143   TUMOR OR CANCER OR CARCINOMA
S7         86   S5 NOT S6
S8         62   RD (unique items)
S9     330155   (NORMAL OR HEALTHY)(5N)(INDIVIDUAL?? OR PERSON?? OR MALE??
             OR SUBJECT??)
S10       105   S9 AND S3
S11         6   S10 NOT S6
S12         4   RD (unique items)
? s circulation or blood
Processing
         1058705  CIRCULATION
         3385902  BLOOD
     S13 3769829  CIRCULATION OR BLOOD
? s s1 and s13
           18595  S1
         3769829  S13
     S14    1832  S1 AND S13
? s s14 and s9
            1832  S14
          330155  S9
     S15      70  S14 AND S9
? rd
>>>Duplicate detection is not supported for File 340.

>>>Records from unsupported files will be retained in the RD set.
...examined 50 records  (50)
...completed examining records
     S16      40  RD (unique items)
? t s16/3,k,ab/1-20
```

The expression of prostate-specific membrane antigen in peripheral **blood** leukocytes.

Lintula S; Stenman U H

Department of Clinical Chemistry, Helsinki University Central Hospital, Finland.

Journal of urology (UNITED STATES) May 1997, 157 (5) p1969-72, ISSN 0022-5347 Journal Code: 0376374

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

PURPOSE: Prostate-specific membrane antigen (PSM) and prostate-specific antigen (PSA) have been used as marker genes for detection of cancer cells in **circulation** of prostate cancer patients. However, PSA was recently found to be expressed in non-**prostate cell** lines and normal **blood** . To evaluate this phenomenon for PSM, we studied its mRNA expression in non-prostatic cells and cell lines, in **blood** from healthy donors and patients with prostate cancer. MATERIALS AND METHODS: We studied PSM expression by a highly sensitive reverse transcription-polymerase chain reaction (RT-PCR) in peripheral **blood** of 24 patients with cancer of the prostate (CAP) and 13 **healthy** young **male** and female donors, in four non-prostatic cell lines and in isolated **blood** cells. The identity of the RT-PCR product was confirmed by sequencing. Contamination of the samples with cDNA or prostatic RNA was rigorously excluded by subjecting each sample to PCR reaction without RT-enzyme and by RT-PCR with PSA primers, respectively. RESULTS: We found PSM mRNA expression in **blood** from 18 of 24 CAP patients and 12 of 13 healthy donors and in the leukocyte fraction of normal **blood** cells. PSM expression could not be detected in erythroblasts, platelets, K-562, U-937, HL-60 or Jurkat cell lines. CONCLUSIONS: Our results indicate that sensitive nested RT-PCR method detects PSM mRNA in the leukocyte fraction of normal **blood**. This "background" expression is probably caused by a leaky promoter of PSM. We conclude that it is necessary to develop quantitative RT-PCR assays to

5/23

3/3,K,AB/19 (Item 19 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

10691871 20576603

Expression of AMPA receptor subunit flip/flop **splice variants** in the rat auditory brainstem and inferior colliculus.

Schmid S; Guthmann A; Ruppersberg JP; Herbert H
Department of Animal Physiology, Institute of Zoology, University of Tubingen, 72076 Tubingen, Germany. susanne.schmid@uni-tuebingen.de

Journal of comparative neurology (UNITED STATES) Feb 5 2001, 430 (2) p160-71, ISSN 0021-9967 Journal Code: HUV
Languages: ENGLISH
Document type: Journal Article

The expression of alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionate (AMPA) receptor subunit mRNAs and their flip/flop **splice variants** was evaluated in the rat auditory brainstem and inferior colliculus employing in situ hybridization with radiolabeled oligonucleotide probes. A **differential expression** of AMPA receptor subunits in auditory nuclei was observed. In general, neurons in all nuclei of the auditory brainstem express high levels of GluR-C flop and GluR-D flop mRNA, but low to very low levels of GluR-A and GluR-B mRNA. The strongest GluR-C and -D flop expression is found in the ventral and medial part of the anteroventral cochlear nucleus, the posteroventral cochlear nucleus, and the medial and the lateral superior olive. These nuclei are part of the binaural auditory pathway which is important for sound localization in space. In contrast, neurons in the central nucleus of the inferior colliculus express high levels of GluR-B flip but only low levels of the other AMPA receptor subunits. From our data, we conclude that neurons of nuclei involved in binaural processing exhibit a specific "auditory AMPA receptor" which consists primarily of GluR-C flop and -D flop and often lacks GluR-B subunits; this indicates fast kinetics and high Ca(2+) permeability of AMPA receptor currents. In contrast, neurons in the central nucleus of the inferior colliculus contain large amounts of GluR-B flip subunits resulting in Ca(2+) impermeable AMPA receptors with slow kinetics. Copyright 2001 Wiley-Liss, Inc.

Expression of AMPA receptor subunit flip/flop **splice variants** in the rat auditory brainstem and inferior colliculus.

...amino-3-hydroxy-5-methyl-4-isoxazolepropionate (AMPA) receptor subunit mRNAs and their flip/flop **splice variants** was evaluated in the rat auditory brainstem and inferior colliculus employing in situ hybridization with radiolabeled oligonucleotide probes. A

6/3,K,AB/2 (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

09940759 99286090

Expression and androgen regulation of C-CAM cell adhesion molecule isoforms in rat dorsal and ventral **prostate**.

Makarovskiy AN; Pu YS; Lo P; Earley K; Paglia M; Hixson DC; Lin SH
Department of Molecular Pathology, The University of Texas, MD Anderson Cancer Center, Houston 77030, USA.

Oncogene (ENGLAND) May 27 1999, 18 (21) p3252-60, ISSN 0950-9232 Journal Code: ONC
Contract/Grant No.: CA64856, CA, NCI; CA42714, CA, NCI; CA16672, CA, NCI
Languages: ENGLISH
Document type: JOURNAL ARTICLE

C-CAM is an epithelial cell adhesion molecule with two major **splice variants** that differ in the length of the cytoplasmic domain. C-CAM1 (long (L)-form) strongly suppresses the tumorigenicity of human **prostate** carcinoma cells. In contrast, C-CAM2 (short (S)-form) does not exhibit tumor-suppressive activity. In the present study we have investigated the functional significance of L-form and S-form C-CAM in rat **prostate** by examining their **expression** and distribution in **different prostate** lobes and their response to androgen deprivation. RNase protection assays with a probe for both C-CAM isoforms detected high levels of C-CAM messages in the rat dorso-lateral **prostate** (DLP). L- and S-form proteins, localized by indirect immunofluorescence using isoform-specific antipeptide antibodies, were co-expressed on the apical surface of **prostate** epithelial cells in normal DLP. Androgen depletion did not significantly change the steady state levels of C-CAM message and protein expression in the DLP, although there was a change in the pattern of protein expression in these lobes. In contrast, C-CAM isoform messages and proteins were undetectable in normal ventral **prostate** (VP) but increased markedly in this lobe in response to castration, producing isoform ratios similar to those in DLP. These results demonstrate that coordinate expression of C-CAM isoforms is maintained in the VP following androgen depletion and suggest that androgen suppresses C-CAM expression in VP but not in DLP. These results suggest that balanced expression of L- and S-form C-CAM is important for normal **prostate** growth and differentiation.

... and androgen regulation of C-CAM cell adhesion molecule isoforms in rat dorsal and ventral **prostate**.

2/14

10362056 99406642 PMID: 10473985

Detection of prostate-specific antigen- or prostate-specific membrane antigen-positive circulating cells in prostatic cancer patients: clinical implications.

Millon R; Jacqmin D; Muller D; Guillot J; Eber M; Abecassis J

Laboratoire de Biologie Tumorale, Centre Paul Strauss, Hopitaux Universitaires, Strasbourg, France. btumorale@strasbourg.fnclcc.fr

European urology (SWITZERLAND) Oct 1999, 36 (4) p278-85, ISSN 0302-2838 Journal Code: ENM

Languages: ENGLISH

Document type: Journal Article

Record type: Completed

OBJECTIVES: To evaluate the clinical benefit from using circulating prostate-specific antigen (PSA) and prostate-specific membrane antigen (PSM) mRNA detection in prostate cancer staging and in follow-up. METHODS: Nested reverse transcriptase-polymerase chain reaction (RT-PCR) assays were performed on RNA extracted from blood drawn from 56 patients with prostate cancer before any treatment. Additionally, assays were done on posttreatment samples from 50 patients who were followed up by serum PSA level, to determine whether any relationship exists between RT-PCR results and tumor recurrence. The **prostate cell** specificity of assays was evaluated by analysis of 21 **blood** samples from women or cystoprostatectomized men. RESULTS: With PSM RT-PCR assay, good sensitivity and prostate cell specificity could not be attained together, since high PSM mRNA illegitimate expression has been shown in some **healthy** donor bloods. For this reason, only PSA RT-PCR assay was used as a clinical marker. PSA mRNA was detected in peripheral blood of 4 out of 31 patients with clinically localized prostate cancer. It showed no relationship to the pathologic stage, but significant relationship to metastatic status, lymph node involvement and Gleason score. During follow-up, circulating PSA mRNA was detected in 8 out of 17 (47%) patients in treatment failure and in only 1 out of 33 (3%) successfully treated patients, with significant relationship between RT-PCR results and concomitant serum PSA levels. CONCLUSION: Our study reveals no significant advantage to PSA RT-PCR assay (1) in improving the staging of clinically local

Service d'Urologie, Service de Biochimie A, and Service d'Anatomie Pathologique, Hopital Necker, Paris, France.
Journal of urology (UNITED STATES) Jun 2000, 163 (6) p2022-9, ISSN 0022-5347 Journal Code: KC7
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
PURPOSE: To determine whether the presence of **prostate**-derived **cells** in the peripheral **blood circulation** is a marker of **prostate** cancer and to define the clinical impact of the test. MATERIALS AND METHODS: We tested the peripheral blood of 99 patients with prostate adenocarcinoma (PAC), 79 of them undergoing radical prostatectomy, and 92 controls (31 **healthy** volunteers, 50 patients with adenoma and 11 with prostatitis) using a highly controlled procedure including reverse-transcriptase polymerase chain reaction (RT-PCR) targeted to prostate-specific antigen (PSA) mRNA. Patients were followed for 26 +/- 12 (range: 4 to 49) months. Forty tumor tissues were analyzed by immunohistochemistry for expression of p53 and E-cadherin antigens. RESULTS: Thirty three (33%) patients with PAC and 2 (2%) controls scored positive ($p < 0.0001$) for the test. Detection of circulating prostatic cells was associated with development of metastases ($p < 0.001$), with relapse ($p < 0.001$) and with a serum PSA level at diagnosis higher than 15 ng./ml. ($p = 0.009$). The rate of development of metastases according to time was significantly higher in patients who scored positive for the test ($p < 0.04$). In a multivariate analysis, only the RT-PCR test was an independent risk factor associated with relapse (RR: 6.7). Finally, E-cadherin expression was significantly lower in the tumor tissues of positive patients as compared with those who scored negative for the test ($p < 0.01$). CONCLUSIONS: This RT-PCR procedure, performed at diagnosis and with appropriate controls, is a clinically useful assay in evaluating the risk of tumor recurrence after radical prostatectomy in patients with PAC.

PURPOSE: To determine whether the presence of **prostate**-derived **cells** in the peripheral **blood circulation** is a marker of **prostate** cancer and to define the clinical impact of th

11175821 21039921 PMID: 11198272

Detection of **prostate**-specific membrane antigen expressing **cells** in **blood** obtained from renal cancer patients: a potential biomarker of vascular invasion.

de la Taille A; Cao Y; Sawczuk IS; Nozemu T; d'Agati V; McKiernan JM; Bagiella E; Buttyan R; Burchardt M; Olsson CA; Bander N; Katz AE

Squier Urological Clinic and Department of Urology, College of Physicians and Surgeons of Columbia University, New York, NY, USA.

Cancer detection and prevention (United States) 2000, 24 (6) p579-88 , ISSN 0361-090X Journal Code: CNZ

Contract/Grant No.: CA 70769, CA, NCI

Languages: ENGLISH

Document type: Evaluation Studies; Journal Article

Record type: Completed

Originally, prostate-specific membrane antigen (PSMA) was described in benign and malignant prostate cells. On the basis of recent reports that this antigen also is expressed in **normal** renal proximal tubular cells and in the neovascular endothelium associated with renal carcinoma, we used a nested reverse transcriptase-polymerase chain reaction assay to evaluate whether PSMA-expressing cells might be present in specimens of peripheral blood obtained from renal cancer patients, benign renal tumor patients, and **healthy** volunteers. Our reverse transcriptase-polymerase chain reaction PSMA assay had a sensitivity of detecting 1 lymph node prostate cancer (LNCaP) per 10(7) lymphocytes. None of the 20 non-renal cancer controls were positive for PSMA mRNA, whereas 11 of 50 patients (22%) with diagnosed renal cancer were positive. Despite a comparative increase of PSMA positivity with stage, no statistical correlation was found. However, 44% of PSMA-positive patients had tumor size greater than 12 cm, versus only 9% in patients negative for PSMA (P = .03), and 67% of positive PSMA patients were found to have vascular invasion versus only 16% of patients negative for PSMA (P = .006; odds ratio, 10.8). This preliminary study suggests the possibility that PSMA expression in peripheral blood might be a useful biomarker for detecting or monitoring the progression of renal cancer in patients

93 (22) p1747-52, ISSN 0027-8874 Journal Code: 7503089
Contract/Grant No.: PO1CA58184, CA, NCI; RO1CA77664, CA, NCI; RO1DE012488, DE, NIDCR; UO1CA84986, CA, NCI
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
BACKGROUND: Methylation of regulatory sequences near GSTP1, which encodes the pi class glutathione S-transferase, is the most common epigenetic alteration associated with prostate cancer. We determined whether the quantitation of GSTP1 methylation in histopathologically distinct prostate tissue samples could improve prostate cancer detection. METHODS: We used a fluorogenic real-time methylation-specific polymerase chain reaction (MSP) assay to analyze cytidine methylation in the GSTP1 promoter in prostate tissue samples from 69 patients with early-stage prostatic adenocarcinoma (28 of whom also had prostatic intraepithelial neoplasia lesions) and 31 patients with benign prostatic hyperplasia. The relative level of methylated GSTP1 DNA in each sample was determined as the ratio of MSP-amplified GSTP1 to MYOD1, a reference gene. We also performed a prospective, blinded investigation to quantitate GSTP1 promoter methylation in sextant prostate biopsy specimens from 21 additional patients with elevated serum prostate-specific antigen levels, 11 of whom had histologically identified adenocarcinoma and 10 of whom had no morphologic evidence of adenocarcinoma. All data were analyzed by using nonparametric two-sided statistical tests. RESULTS: The median ratios (and interquartile ranges) of MSP-amplified GSTP1 to MYOD1 in resected benign hyperplastic prostatic tissue, intraepithelial neoplasia, and adenocarcinoma were 0 (range, 0-0.1), 1.4 (range, 0- 45.9), and 250.8 (range, 53.5-697.5), respectively; all of these values were statistically significantly different (P< .001). The median ratios of MSP-amplified GSTP1 to MYOD1 in the prospectively collected sextant biopsy samples were 410.6 for the patients with adenocarcinoma and 0.0 for the patients with no evidence of adenocarcinoma (P< .001). CONCLUSION: Quantitation of GSTP1 methylation accurately discriminates between **normal** hyperplastic tissue and prostatic carcinoma in small samples of prostate tissue and may augment the standard pathologic/histologic assessment of the prostate.

...with no evidence of adenocarcinoma (P< .001). CONCLUSION: Quantitation of GSTP1 methylation accurately discriminates between **normal** hyperplastic tissue and prostatic carcinoma in small samples of prostate tissue and may augment the...
; Cytidine--metabolism--ME; Fluorescence; Gene Expression; Polymerase Chain Reaction; Promoter Regions (Genetics)--genetics--GE; **Prostate**--pathology--PA; **Prostate**-Specific Antigen--**blood**--BL; **Prostate**-Specific Antigen--metabolism--ME; Prostatic Neoplasms--pathology--PA; Tumor **Cells**, Cultured

12/3,K,AB/2 (Item 2 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

12514261 21327239 PMID: 11434386
Real-Time quantitative reverse transcriptase-polymerase chain reaction (RT-PCR) for the measurement of prostate-specific antigen mRNA in the peripheral blood of patients with prostate carcinoma using the taqman detection system.
Gelmini S; Tricarico C; Vona G; Livi L; Melina A D; Serni S; Cellai E; Magrini S; Villari D; Carini M; Serio M; Forti G; Pazzagli M; Orlando C
Clinical Biochemistry Unit, University of Florence, Italy.
Clinical chemistry and laboratory medicine : CCLM / FESCC (Germany) May 2001, 39 (5) p385-91, ISSN 1434-6621 Journal Code: 9806306
Languages: ENGLISH
Document type: Journal Article
Record type: Completed
Circulating **prostate cells** can be detected in peripheral

2/14

blood of patients with clinically localized or advanced **prostate** carcinoma. Traditionally, nested reverse transcriptase-polymerase chain reaction (RT-PCR) is used for this as a sensitive, but qualitative only, detection system. We developed a quantitative real-time RT-PCR method for measuring prostate-specific antigen (PSA) mRNA in peripheral blood of prostate cancer patients. A quantitative assay was developed using an external standard reference curve generated with RNA from the human **prostate cell** line LNCaP. Basal **blood** samples were collected from 44 patients without evidence of distant metastases and from 30 **healthy** controls. In 29 patients surgically treated with radical prostatectomy, the measurement of PSA mRNA was performed in blood samples collected before, at the end and 6 days after surgery. In 14 patients treated with radiotherapy, the measurements were repeated at 3-month intervals to evaluate time-related changes during therapy. The measurements were also performed for one year at 3-month intervals in one patient treated with anti-androgen therapy. We found detectable PSA mRNA in 14/44 (32%) basal blood samples. A wide range of values were observed in these patients, ranging from 0.5 to 1724 pg of total LNCaP RNA/ml blood. In patients undergoing radical prostatectomy, circulating PSA mRNA was detectable in eight patients in basal samples, and in seven of them also in blood specimens collected at the end of surgery, showing an increase in only two patients. In blood samples collected 6 days later, PSA mRNA was dramatically reduced in all patients, but still present in seven of them. In four patients, whose basal samples were negative, PSA mRNA was detectable in samples collected at the end of surgery and three of them were negative after 6 days. In patients who did not receive surgical treatment, a rapid decrease in PSA mRNA was demonstrated in five patients treated with radiotherapy and in one patient undergoing androgen deprivation. No detectable PSA mRNA was found in **healthy** controls. The levels of PSA mRNA in peripheral blood from patients with prostate carcinoma can be easily measured by this sensitive, quantitative and reliable procedure. This assay is a promising tool for the detection and follow-up of these patients.

Circulating **prostate cells** can be detected in peripheral **blood** of patients with clinically localized or advanced **prostate** carcinoma. Traditionally, nested reverse transcriptase-polymerase chain reaction (RT-PCR) is used for this as...

...assay was developed using an external standard reference curve generated with RNA from the human **prostate cell** line LNCaP. Basal **blood** samples were collected from 44 patients without evidence of distant metastases and from 30 **healthy** controls. In 29 patients surgically treated with radical prostatectomy, the measurement of PSA mRNA

Quantitative polymerase chain reaction does not improve preoperative prostate cancer staging: a clinicopathological molecular analysis of 121 patients.

Sokoloff MH; Tso CL; Kaboo R; Nelson S; Ko J; Dorey F; Figlin RA; Pang S; deKernion J; Belldegrun A

Department of Surgery, UCLA School of Medicine 90095-1738, USA.

Journal of urology (UNITED STATES) Nov 1996, 156 (5) p1560-6, ISSN 0022-5347 Journal Code: KC7

Comment in J Urol. 1998 Apr;159(4) 1311; Comment in J Urol. 1998 Apr;159(4):1311-3

Languages: ENGLISH

Document type: Clinical Trial; Journal Article

Record type: Completed

2/14

PURPOSE: To improve on current staging and monitoring methods for prostate cancer, we applied the technique of quantitative polymerase chain reaction to measure the degree of tumor burden in the circulation and correlate this with pathological tumor stage. A reproducible, highly sensitive and specific, reverse transcriptase-polymerase chain reaction amplification technique to quantify prostate specific antigen (PSA) and prostate specific membrane antigen gene expression in the peripheral circulation was developed. Using a 32phosphorus-gamma-adenosine triphosphate-5'PSA and prostate specific membrane antigen primer incorporation assay, the ribonucleic acid signal extracted from a single neoplastic **cell** (LNCaP) premixed in 10 cc **normal** whole **blood** could be amplified. PSA and **prostate** specific membrane antigen polymerase chain reaction indexes have been created for clinical application. MATERIALS AND METHODS: From September 1994 through July 1995 specimens from 121 patients were prospectively analyzed for PSA and prostate specific membrane antigen signals. RESULTS: Circulating PSA producing cells were present in 29 of 33 patients (88%) with metastatic prostate cancer. Two of 19 patients (11%) with no known prostate cancer exhibited positive signals (1 later had prostate cancer), establishing a sensitivity of 88% and specificity of 94% for our assay. Positive PSA polymerase chain reaction signals were detected in 30 of 51 patients (59%) with stages pT1 and pT2 disease and in 13 of 18 (72%) with stage pT3 cancer. No statistically significant relationship of a positive PSA polymerase chain reaction signal to pathological stage, tumor grade, apical involvement or positive surgical margins was found, and no benefit was derived by measuring the quantity of circulating PSA polymerase chain reaction signals. Circulating prostate specific membrane antigen polymerase chain reaction signals were identified mostly in patients with advanced prostate cancer and offered no benefit to preoperative staging. CONCLUSIONS: Given the high incidence of false positive signals in patients with pathologically determined localized disease, in our experience polymerase chain reaction based assays offer no immediate benefit for preoperative prostate cancer staging. The prognostic significance of detecting circulating prostate specific signals awaits longer followup in this cohort of patients, which is currently under study.

... specific membrane antigen primer incorporation assay, the ribonucleic acid signal extracted from a single neoplastic **cell** (LNCaP) premixed in 10 cc **normal** whole **blood** could be amplified. PSA and **prostate** specific membrane antigen polymerase chain reaction indexes have been created for clinical application. MATERIALS AND...

14/3,K,AB/16 (Item 16 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

07961325 94062397 PMID: 8243118

Prevalence of human papillomaviruses 16 and 18 in transitional cell carcinoma of bladder.

Yu ST; Wu MM; Li LM

Department of Urolog

323167 98331136 PMID: 9666686

[Detection of PSA mRNA from the peripheral blood and pelvic lymph nodes in patients with prostatic cancer by means of reverse transcription-polymerase chain reaction (RT-PCR)]

Mao H; Hoshi S; Takahashi T; Kaneda T; Wang J; Suzuki K; Orikasa S
Department of Urology, Tohoku University School of Medicine, Sendai.

Nippon Hinyokika Gakkai zasshi (JAPAN) Jun 1998, 89 (6) p596-603, ISSN 0021-5287 Journal Code: KRB
Languages: JAPANESE
Document type: Journal Article
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BACKGROUND AND METHODS: To detect prostate cancer cells in the blood circulation and in the lymph nodes by RT-PCR methods, we examined two kinds of prostate specific antigens (PSA) primers and one prostate specific membrane antigen (PSM). PSa primer 1 was established by us, PSA primer 2 by Moreno et al and PSM primer by Israeli et al. RESULTS: Both PSA primers were specific for expression of PSA mRNA because in 12 kinds of urogenital culture cells only LNCaP cells, which produce PSA, expressed PSA mRNA by RT-PCR, PSA 1 was more sensitive than PSA 2 for detection of PSA mRNA in the circulating cells since PSA mRNA was detected in the blood circulating cells in 5 cases of stage D2 prostate cancer using PSA primer 1 but in only one was using PSA primer 2. PSM mRNA was detected in all 12 types of urogenital cancer **cells** and in the **blood** circulating **cells** not of **prostate** cancer patients but also of renal, bladder, testicular cancer patients and **normal** volunteers. PSA 1 was used to detect PSA mRNA from the samples of fine needle aspiration biopsy (FNAB) of pelvic lymph node, and PSA mRNA was positive in 10 FNAB samples including not only all 6 cytologically positive and two cytologically class III cases but also 2 of 8 cytologically negative cases. RT-PCR for FNAB samples of all 15 cases of bladder cancer were negative for the detection of PSA mRNA. CONCLUSION: Detection of PSA mRNA by RT-PCR in FNAB samples may be useful to diagnose pelvic lymph node metastasis and to furnish additional information for the cytological diagnosis of prostate cancer.

... using PSA primer 2. PSM mRNA was detected in all 12 types of urogenital cancer **cells** and in the **blood** circulating

0312037 97266850 PMID: 9112573
The expression of prostate-specific membrane antigen in peripheral blood leukocytes.
Lintula S; Stenman UH
Department of Clinical Chemistry, Helsinki University Central Hospital, Finland.
Journal of urology (UNITED STATES) May 1997, 157 (5) p1969-72, ISSN 0022-5347 Journal Code: KC7
Languages: ENGLISH
Document type: Journal Article
Record type: Completed

2/14

PURPOSE: Prostate-specific membrane antigen (PSM) and prostate-specific antigen (PSA) have been used as marker genes for detection of cancer **cells** in **circulation** of **prostate** cancer patients. However, PSA was recently found to be expressed in non-**prostate cell** lines and **normal blood**. To evaluate this phenomenon for PSM, we studied its mRNA expression in non-prostatic **cells** and cell lines, in blood from healthy donors and patients with prostate cancer. MATERIALS AND METHODS: We studied PSM expression by a highly sensitive reverse transcription-polymerase chain reaction (RT-PCR) in peripheral blood of 24 patients with cancer of the prostate (CAP) and 13 healthy young male and female donors, in four non-prostatic cell lines and in isolated blood cells. The identity of the RT-PCR product was confirmed by sequencing. Contamination of the samples with cDNA or prostatic RNA was rigorously excluded by subjecting each sample to PCR reaction without RT-enzyme and by RT-PCR with PSA primers, respectively. RESULTS: We found PSM mRNA expression in blood from 18 of 24 CAP patients and 12 of 13 healthy donors and in the leukocyte fraction of normal blood cells. PSM expression could not be detected in erythroblasts, platelets, K-562, U-937, HL-60 or Jurkat cell lines. CONCLUSIONS: Our results indicate that sensitive nested RT-PCR method detects PSM mRNA in the leukocyte fraction of normal blood. This "background" expression is probably caused by a leaky promoter of PSM. We conclude that it is necessary to develop quantitative RT-PCR assays to differentiate PSM mRNA expression derived from circulating cancer cells from background expression in blood cells.

...and prostate-specific antigen (PSA) have been used as marker genes for detection of cancer **cells** in **circulation** of **prostate** cancer patients. However, PSA was recently found to be expressed in non-**prostate cell** lines and **normal blood**. To evaluate this phenomenon for PSM, we studied its mRNA expression in non-prostatic **cells** and cell lines, in blood from healthy donors and patients with prostate cancer. MATERIALS AND...

14/3,K,AB/8 (Item 8 from file: 155)
DIALOG(R)File 155:MEDLINE(R)

10240912 99362750 PMID: 10430935
High expression of a specific T-cell receptor gamma transcript in epithelial cells of the prostate.
Essand M; Vasmatzis G; Brinkmann U; Duray P; Lee B; Pastan I
Laboratory of Molecular Biology, Division of Basic Sciences, Division of Clinical Sciences, National Cancer Institute, National Institutes of Health, Building 37, Room 4E16, Bethesda, MD 20892, USA.
Proceedings of the National Academy of Sciences of the United States of America (UNITED STATES) Aug 3 1999, 96 (16) p9287-92, ISSN 0027-8424 Journal Code: PV3
Languages: ENGLISH
Document type: Journal Article
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We have identified expression of T-cell receptor gamma chain (TCRgamma) mRNA in human prostate and have shown that it originates from epithelial cells of the prostate and not from infiltrating T-lymphocytes. In contrast,

the T-cell receptor delta chain (TCRdelta) gene is silent in human prostate. The major TCRgamma transcript in prostate has a different size than the transcript expressed in thymus, spleen, and **blood** leukocytes. It is expressed in **normal prostate** epithelium, adenocarcinoma of the **prostate**, and the prostatic adenocarcinoma **cell** line LNCaP. The RNA originates fr

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? ds

Set      Items   Description
S1       20033   PROSTATE(5N)CELL??
S2     3567842   BLOOD OR CIRCULATION
S3        2003   S1 AND S2
S4      710103   PROGRESS?
S5         280   S3 AND S4
S6     2295387   CANCER? OR TUMOR? OR MALIGNAN?
S7         273   S5 AND S6
S8           0   PROTATE(5N)CELL??(5N) (CIRCULATION OR BLOOD)
S9         495   PROSTATE(5N)CELL??(5N)(CIRCULATION OR BLOOD)
S10    2060337   NORMAL OR HEALTHY
S11        137   S9 AND S10
S12         84   RD (unique items)
? s normal(5n)prostate(5n)cell??(5n)(blood or circulation)

Processing
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           125206  PROSTATE
          5795278  CELL??
          3202880  BLOOD
          1019120  CIRCULATION
     S13       54  NORMAL(5N)PROSTATE(5N)CELL??(5N)(BLOOD OR CIRCULATION)
? rd

>>>Duplicate detection is not supported for File 340.

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...completed examining records
     S14       27  RD (unique items)
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0573710 20096197 PMID: 10632336

Prostate-specific membrane antigen levels in sera from healthy men and patients with benign prostate hyperplasia or prostate cancer.

Beckett M L; Cazares L H; Vlahou A; Schellhammer P F; Wright G L

Department of Microbiology and Molecular Cell Biology, Virginia Prostate Center, Eastern Virginia Medical School, Norfolk 23501, USA.

Clinical cancer research : an official journal of the American Association for Cancer Research (UNITED STATES) Dec 1999, 5 (12) p4034-40, ISSN 1078-0432 Journal Code: 9502500

Contract/Grant No.: CA 26659; CA; NCI; DK47754; DK; NIDDK

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

Prostate-specific membrane antigen (PSMA) serum levels have been proposed to be of prognostic significance in patients with advanced prostate disease. The objective of the present study was to confirm PSMA serum expression by Western blot techniques, to determine whether such data could assist in the differentiation of benign from malignant prostatic disease, and to determine the suitability of serum PSMA measurements in predicting recurrent or progressive prostate malignancies. We measured PSMA, a transmembrane glycoprotein identified in **prostate** epithelial **cells**, in the sera of 236 **normal individuals** and cancer patients by Western blot analysis. Within the **normal male** population, PSMA levels increase with age and were found to be significantly elevated in subjects more than 50 years of age when compared to those of younger men. We did not confirm previous reports that serum PSMA measurements could distinguish late-stage prostate carcinoma from early-stage prostate carcinoma, nor did we find PSMA to be more effective than prostate-specific antigen in monitoring prostate cancer patient prognosis. Furthermore, we found elevated serum PSMA in healthy females, and, similar to the **healthy male** population, the levels increased with age, with the highest levels found in the sera from breast cancer patients. These latter observations further support that PSMA is not a specific biomarker for prostate cancer and that a variety of normal and diseased tissue may contribute to the serum levels of PSMA.

1072058 21099575 PMID: 11173941

Blood -borne RT-PCR assay for prostasin- specific transcripts to identify circulating **prostate cells** in cancer patients.

Laribi A; Berteau P; Gala J; Eschwege P; Benoit G; Tombal B; Schmitt F; Loric S

Biochemistry A Laboratory, Saint-Antoine AP-HP University Hospital, Paris, France.

European urology (Switzerland) Jan 2001, 39 (1) p65-71, ISSN 0302-2838 Journal Code: 7512719

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: Completed

OBJECTIVE: The aim of this study was to establish the specific detection of prostasin-expressing **prostate cells** in the **blood** of patients with prostate cancer. PATIENTS AND METHODS: A prostasin-specific RT-PCR assay was developed and optimized using limiting dilutions of cell line LNCaP mixed with normal **blood** specimens. Then, it was used to examine peripheral **blood** samples from 96 patients with prostate cancer (localized carcinoma, n = 69, metastatic, n = 27). Specificity was assessed by examination of 86 negative controls (**healthy individuals**, n = 47, benign prostate hyperplasia, n = 17, nonprostate cancer patients, n = 22). RESULTS: All 86 control samples failed to amplify the specific 546-bp prostasin PCR products. **Blood** samples from 35 out of 96 (36%) prostate cancer patients were found positive. In metastatic patients, 63% (17/27) scored positive whereas in localized adenocarcinoma prostasin primers detected **prostate cells** in 26% (18/69). CONCLUSION: Our results that approximately 30% of patients with localized prostate cancer scored positive for prostasin-specific RT-PCR confirm that the hematogenous spillage of **prostate cells** is an early event in the natural history of prostate cancer. As none of our negative controls were found positive, we conclude that **blood** -borne RT-PCR amplification of prostasin transcripts may lead to an earlier diagnosis of disseminated disease in patients with organ-confined carcinoma. The clinical significance of **prostate cell** detection and the

The clinical utility of the prostate specific membrane antigen reverse-transcription/polymerase chain reaction to detect circulating **prostate cells**: an analysis in healthy men and women.

Llanes L; Ferruelo A; Paez A; Gomez J M; Moreno A; Berenguer A
Department of Urology, Hospital Universitario de Getafe, Madrid, Spain.

BJU international (England) Jun 2002, 89 (9) p882-5, ISSN 1464-4096
Journal Code: 100886721
Document type: Journal Article
Languages: ENGLISH
Main Citation Owner: NLM
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OBJECTIVE: To evaluate the overall specificity of nested reverse transcriptase-polymerase chain reaction (RT-PCR) to detect prostate-specific membrane antigen (PSM) mRNA in peripheral **blood** samples of **healthy** donors. **SUBJECTS** AND METHODS: Peripheral **blood** samples were taken from 60 healthy **blood**-donors (30 men and 30 women aged < 50 years) and analysed for PSM-mRNA using nested RT-PCR (in 'hot-start' conditions and confirmed using nested EcoRI restriction enzyme). Intron-spanning primer pairs specific for human PSM were deduced from the GenBank sequence (M99487) using gene software. The outer primer pair for PSM was: fwd: 1368 5'-TCACCGGGACTCATGGGTGT-3'; reverse: 1860 5'-GCCTGAAGCAATTCCAAGTCGG-3'. Inner primer pair for PSM was: fwd: 1480 5'-AAGGAAGGGTGGAGACCTAG-3'; reverse: 5-ACTGAACTCTGGGGAAGGAC-3'. The integrity of cDNAs was checked using primer pairs specific for the housekeeping gene beta-actin. The specificity and false-positive rate were calculated assuming that the underlying prostate cancer incidence was nil. RESULTS: The first PCR was negative for all samples (100% specificity; 0% false-positive rate). The nested PCR detected 23 positive samples (23/60, 38%) with an overall specificity of 62% (false positive rate, 38%). CONCLUSION: Nested RT-PCR of PSM-mRNA in peripheral **blood** is highly unspecific. Its clinical utility in the management of prostate cancer must be low. Further development is needed of quantitative RT-PCR, primers that identify prostatic PSM or another prostate-specific marker gene to differentiate PSM mRNA from circulating **prostate cells** and from

706252 22260204 PMID: 12373303

Detection of circulating **prostate** tumor **cells**: alternative spliced variant of PSM induced false-positive result.

Hisatomi H; Nagao K; Kawakita M; Matsuda T; Hirata H; Yamamoto S; Nakamoto T; Harasawa H; Kaneko N; Hikiji K; Tsukada Y

Center for Molecular Biology and Cytogenetics, SRL, Inc., Hino, Tokyo 191-0002, Japan. hisatomi@srl.srl-inc.co.jp

International journal of molecular medicine (Greece) Nov 2002, 10 (5) p619-22, ISSN 1107-3756 Journal Code: 9810955

Document type: Journal Article

Languages: ENGLISH

Main Citation Owner: NLM

Record type: In Process

RT-nested PCR has been introduced as a highly specific and sensitive assay method to detect the prostate-specific membrane antigen (PSM) mRNA in peripheral **blood**. However, appreciable percentages of false-positive cases have been reported. Additionally, primer sets reported previously could not discriminate between PSM and PSM', an alternatively spliced variant, mRNA. These isoforms can be produced from a single gene. Switches in alternative splicing patterns are often controlled with strict cell-type or developmental-stage specificity. Therefore, it is most important to discriminate between PSM mRNA and PSM' mRNA. Using our highly specific primer sets, PSM mRNA was detected in 3 of 24 peripheral **blood** samples of **normal male** volunteers (12.5%) and was not detected in peripheral **blood** of 11 normal female volunteers. PSM' mRNA was detected in 5 of 24 peripheral **blood** samples of **normal male** volunteers (20.8%) and in 4 of 11 of normal female volunteers (36.4%). PSM' mRNA induced false-positive results, it is important for genetic diagnosis of prostate cancer to discriminate between PSM and PSM' using our primer sets with high specificity. The advances in the uniquely designed primer sets may allow researchers to detect a real PSM mRNA without PSM' mRNA.